

WHAT IS CLAIMED IS:

1. A method of making didanosine (ddI) comprising the steps of:
 - (a) obtaining an enzyme expressing ddA deaminase activity;
 - 5 (b) immobilizing the enzyme onto an insoluble support;
 - (c) contacting the enzyme with a dideoxyadenosine (ddA) solution of at least about 1% weight volume ddA in water for a time and under conditions to produce a ddI solution; and
 - (d) isolating the ddI from the ddI solution.
- 10 2. The method of claim 1, wherein the ddA solution in the contacting step is from about 2% to about 10% weight volume ddA in water.
3. The method of claim 1, wherein a pH during the contacting step is from about
15 8.0 to about 9.5.
4. The method of claim 3, wherein substantially all of the ddI resists precipitation out of the ddI solution in the contacting step.
- 20 5. The method of claim 1, wherein the insoluble support is functionalized to allow attachment of the enzyme thereto.
6. The method of claim 5, wherein the attachment of the enzyme to the insoluble support is achieved using an activating agent.
- 25 7. The method according to claim 1, wherein the enzyme is human adenosine deaminase (ADA).
8. The method of claim 7, wherein the ADA has the amino acid sequence of SEQ
30 ID NO:1, or conservative variations thereof.
9. The method of claim 7, wherein the ADA is coded for by a nucleotide having SEQ ID NO: 2, SEQ ID NO:3, or conservative variations thereof.

10. The method according to claim 1, wherein the obtaining step includes expressing human adenosine deaminase (ADA) or a conservative variant thereof in a transformed organism and isolating the ADA from the organism.
- 5 11. The method of claim 10, wherein the transformed organism is *E. coli*.
12. The method of claim 10, wherein the insoluble support is functionalized to allow attachment of the enzyme thereto.
- 10 13. The method of claim 12, wherein attachment of the enzyme to the insoluble support is achieved using an activating agent.
14. The method of claim 10, wherein an activity of the enzyme immobilized on the insoluble support is at least about 40 U/g.
- 15 15. The method of claim 10, wherein a pH during the contacting step is from about 7.5 to about 9.5.
16. The method of claim 10, wherein said contacting step is a continuous process performed using a packed column.
- 20 17. The method of claim 10, wherein the ddA solution in the contacting step is from about 4% to about 15% weight volume ddA in water.
- 25 18. The method of claim 17, wherein the ddA solution is from about 5% to about 8% weight volume ddA in water.
19. The method of claim 10, wherein the isolating step includes sequentially distilling the ddI solution and adding water until a ddI slurry in aqueous mother liquor is obtained and the pH is less than about 8.
- 30 20. The method of claim 10, further comprising the steps of:

- (a) retaining a reaction mother liquor after the isolating step; and
- (b) repeating the contacting step at least once using the reaction mother liquor to prepare the ddA solution; and
- (c) repeating the isolating step at least once.

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21. The method of claim 20, wherein the isolating step produces a yield of at least about 96% ddI that is at least about 99% pure.